



**UNITED STATES ENVIRONMENTAL PROTECTION AGENCY  
REGION 8**

1595 Wynkoop Street  
DENVER, CO 80202-1129  
Phone 800-227-8917  
<http://www.epa.gov/region08>

Ref: 8EPR-SR

March 16, 2009

MEMORANDUM

SUBJECT: Coordination with the Libby OU3 Biological Technical Assistance Group on the Design of a Surface Water Toxicity Study, Phase III Remedial Investigation

FROM: Bonnie Lavelle  
Remedial Project Manager

TO: Libby Asbestos Site OU3 Site File

EPA Region 8 consulted with the Libby OU3 Biological Technical Assistance Group (BTAG) on the proposed design of a surface water toxicity study using rainbow trout to support the OU3 ecological risk assessment.

EPA Region 8 provided the attached proposed Fiber Pilot Study Design (Attachment 1) and characteristics of the proposed spiking material (Attachment 2) to BTAG members on February 19, 2009. EPA Region 8 conducted a conference call with the BTAG to discuss the design on March 2, 2009.

The following BTAG members participated in the March 2, 2009 conference call:

Dan Wall, US Fish and Wildlife Service  
Karen Nelson, US Fish and Wildlife Service  
Richard Henry, US Fish and Wildlife Service  
David Charters, EPA ERT  
Bob Marriam, Remedium  
Bob Medler, Remedium  
Bill Stubblefield, Parametrix  
Sue Robinson, Parametrix  
Bill Brattin, SRC  
Janet Burris, SRC  
Bonnie Lavelle, EPA Region 8

The main items of discussion during the March 2, 2009 conference call were:

- There was consensus that a spiking study using a flow-through design is preferred and

that the available information on fiber size distribution of potential spiking material sufficiently matches the fiber size distribution of LA in surface water collected at OU3 for use in the study.

- It's unknown whether there's enough spiking material currently available to support the OU3 study. EPA Region 8 will work with USGS to obtain spiking material and information on its characteristics. It's possible that the OU3 study will have to use material that is currently being processed and characterized by USGS. EPA Region 8 will work with USGS to determine when this material will be available.
- Parametrix suggested using 5 dilutions in the full study rather than the 6 proposed.
- EPA Region 8 will consult with the analytical laboratories to make the final determination on the analytical requirements.
- The BTAG discussed the appropriate water concentration to test in the study. Some BTAG members questioned whether it's appropriate to use a water concentration that represents a short term high concentration in a chronic toxicity study. The alternative would be to use a concentration that characterizes an average concentration within an exposure unit. They also questioned whether it's appropriate to use a concentration that was measured in a pond at the site if the pond is not good fish breeding habitat. EPA Region 8 will provide more detailed information on how the results of the study will be used in the risk assessment and risk management in order to support the choice of the water concentration to be tested.

Attachment 3 is an analysis of the amount of spiking material needed for the study under several alternative designs.

## Attachments

Bonita  
Lavelle/EPR/R8/USEPA/US

02/19/2009 06:21 PM

To Dan Wall/EPR/R8/USEPA/US@EPA, Davidw  
Charters/ERT/R2/USEPA/US@EPA, Richard  
Henry/ERT/R2/USEPA/US@EPA, Karen\_Nelson@fws.gov,  
cc Wendy OBrien/EPR/R8/USEPA/US@EPA, Mary  
Goldade/EPR/R8/USEPA/US@EPA

bcc

Subject fish tox test pilot study design

Hi all

Attached please find a proposed design for a pilot study as we discussed at the BTAG meeting on Feb 4.

Depending on your availability, I'd like to have a conference call with the BTAG to discuss your comments either Friday February 27 or Monday March 2.

The goal is to incorporate the final pilot study design into the final Phase III Sampling and Analysis Plan which I hope to complete by early April.

Please let me know if you are available for a call. I suggest 10 AM mountain time since we have folks participating from all time zones.



Fiber Pilot Study v2.doc USGS Dirty 6 vs L4 in SW OU3\_Histogram.pdf

Sincerely,

Bonnie Lavelle  
Remedial Project Manager  
Libby Asbestos Superfund Site, OU3  
EPA Region 8  
1595 Wynkoop Street  
8EPR-SR  
Denver, CO 80202-1129

(303) 312-6579  
Fax (303) 312-7151

ATTACHMENT 1

## **LIBBY OU3 FIBER PILOT STUDY DESIGN**

### **1.0 INTRODUCTION**

- A fish toxicity test using OU3 site water was previously performed.
- Measures of LA concentration indicate that fibers were being lost from the carboy and the aquaria during the test.
- The reason is not certain, but one plausible explanation is time-dependent formation of a bio-film on the walls of the carboy and aquaria, which in turn adsorbed free fibers from the test water
- The purpose of this pilot study is to find a way to ensure that a repeat toxicity test will not be hampered by the same problem

### **2.0 REVISED TOX STUDY DESIGN**

#### Spiking Study vs Site Water

Two basic options are available for performing a fish toxicity test:

- Using authentic site water as test medium
- Using lab water spiked with LA

Each approach has some pros and some cons.

In general, a spiking design is easier to implement and control than a site water test, and it is likely to be less vulnerable to bio-film effects than a site water test. Thus, this is the design preferred by EPA. However, this is conditional on the assumption that the properties of LA in the spiking material are similar to that of LA which is seen in site waters.

Sufficient data are now available to characterize the particle size distribution in site water, and this can be compared to the size distribution in one potential spiking material (USGS "Dirty 6"). See Figure 1. These data suggest the length distribution is very well matched, although LA particles in site water tend to be somewhat thinner than in the potential spiking material.

USGS is presently preparing a new batch of LA that is intended to be generally similar to the "Dirty 6". Once prepared, USGS will characterize the particle size distribution of the new material, and this can be compared to LA in OU3 site water.

EPA Region 8 is proposing that a decision regarding the spiking material be deferred until the new material is characterized. If the new material matches OU3 site water as well or better than the "Dirty 6" material, then this new material will be used for spiking. If the new material has a particle size distribution that matches less well than the "Dirty 6", then EPA proposes using the "Dirty 6" material.

### Flow-Through Design

The previous test using site water employed a static renewal design. One advantage of a spiking design is that the test can be run using continual flow-through. This will help ensure that buildup of ammonia or other waste products in the water is minimized, and it may also help minimize bio-film growth problems. For this reason, a flow-through design is planned for the toxicity study.

The highest concentration to be tested will be at least 30 MFL, and potentially higher, subject to input from the BTAG. Lower concentrations will be prepared by serial 1/10 dilutions, such that the relative concentrations are:

- 100%
- 10%
- 1.0%
- 0.1%
- 0.01%
- 0.01%
- Zero (un-spiked lab water)

Each concentration will be tested in quadruplicate (i.e., 4 aquaria per concentration level).

Test organisms will be rainbow trout fry. Exposure will last 42 days, including about 20 days as sack fry and about 21 days post swim-up.

Samples of water will be collected from the aquaria over time during the test and analyzed for fibers by both PCM (real time but screening level) and TEM (slower but definitive) to ensure that target fiber concentrations are being achieved.

### **3.0 Pilot Study Design**

The purpose of the pilot study is to determine, if the toxicity test were performed as described above, if the concentration of LA fibers in test chambers would be as expected, or would fiber loss occur (and when).

In order to achieve this goal, it is necessary that the pilot study mimic all of the key features of the main design, as described above.

Based on this, the design of the pilot study is as follows:

- 1) Set up the test system exactly as will be done during the authentic test.
- 2) In one set of aquaria, place no organisms
- 3) In the second set of aquaria, place sac fry
- 4) In the third set of aquaria, place fry that are 20+ days post swim-up. Feed as usual
- 5) Begin the flow-through exposure on day 1, and continue for 21 days

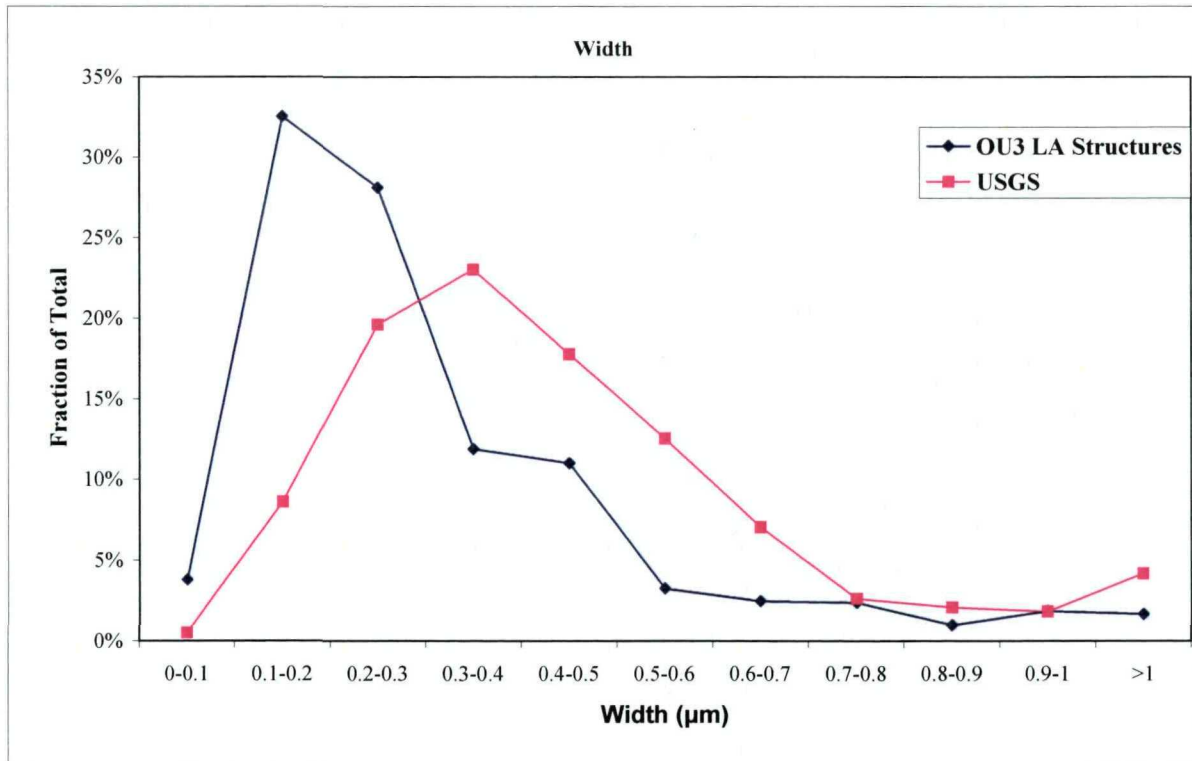
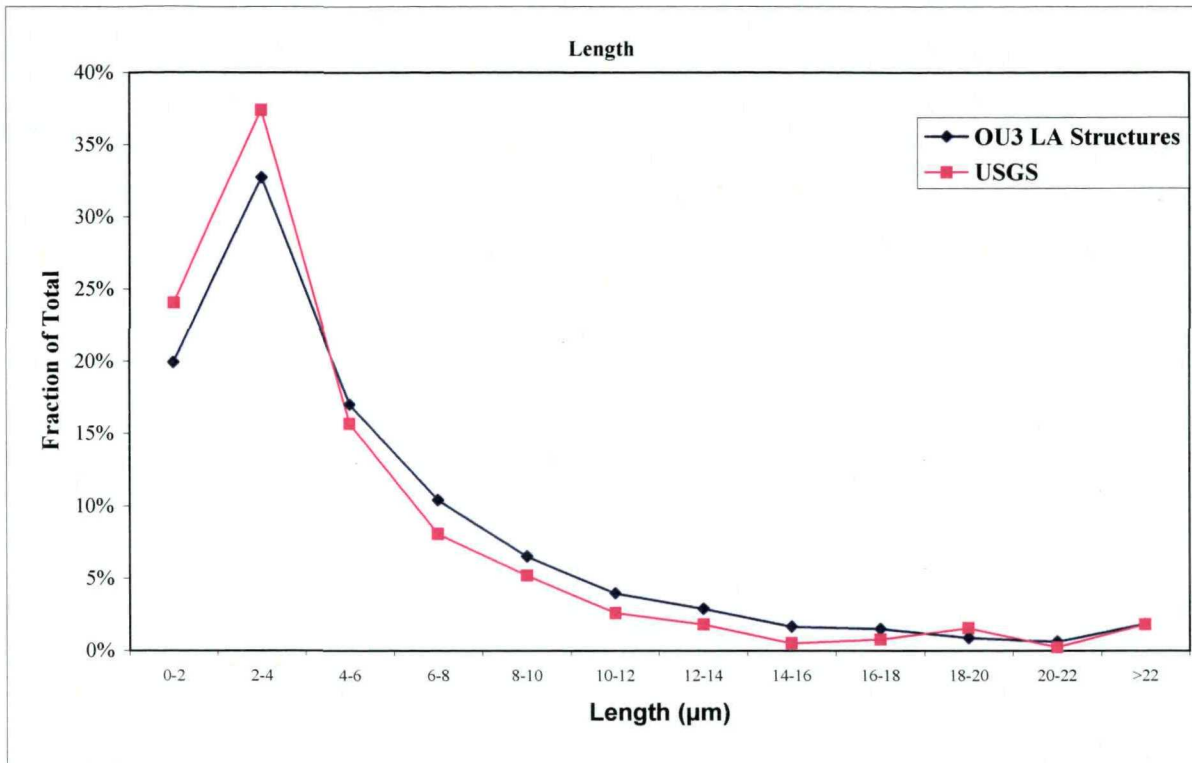
- 6) Withdraw a sample of water from the stock water bottle (5 mL) and from the 100% (5 mL), 1% (50 mL) and 0.01% (250 mL) dilutions in each series (no organisms, sac fry, or swim-up) on days 1, 3, 5, 10, 15 and 21. This corresponds to a total of **60** samples of water for analysis.
- 7) Filter each sample through a 385 mm<sup>2</sup> MCE filter and analyze by PCM, counting until 100 fibers or 200 fields of view have been analyzed (whichever comes first). Archive all filters for potential analysis by TEM, as may be judged necessary.

The results of the pilot study will identify the time course of fiber loss (if any) over a 21-day flow-through test. If no loss of fibers is observed in any of the three series (no organisms, sac fry, or swim-up), then the final study design will include a 20 day exposure of sac fry followed by a change out of aquaria and dilutors on day 21, which would then be left in place for the duration of the study.

If fiber loss is noted in any of the test series, then one of 2 options will be pursued:

1. either modify the study design to specify the change out of stock bottle, dilutors and aquaria prior to the time the loss begins, or
2. perform additional pilot studies to further clarify the cause of the fiber loss and identify steps that can prevent the loss

# USGS Dirty 6 vs. OU3 LA Fibers Only



**Estimated Mass (grams) of LA Needed  
for Flow-Through Toxicity Test**

Max C (MFL)	Changes/Day		
	60	30	10
1000	2268	1134	378
100	227	113	38
30	68	34	11

**Assumptions:**

1.5 L/aquarium  
 4 aquaria per concentration  
 42 day study  
 85% Purity  
 8.50E-11 grams per fiber

**PCM Analysis**

C	MFL	100	1.0	0.01
V	mL	5	50	250
N	fibers	500000	50000	2500
EFA	mm2	385	385	385
Loading	f/mm2	1299	130	6
A(FOV)	mm2	0.00785	0.00785	0.00785
f per FOV		10	1.02	0.051
FOVs		10	100	200
Total N		102	102	10

ATTACHMENT 3